In the matter of

State of Oklaholma, ex rel., A. Drew Edmondson in his capacity as Attorney General of the State of Oklahoma, and Oklahoma Secretary of the Environment, C. MILES TOLBERT, in his capacity as the Trustee for Natural Resources for the State of Oklahoma, Plaintiffs

v.

Tyson Foods, Tyson Poultry, Tyson Chicken, Inc., Cobb-Vantress, Inc., Aviagen, Inc., Cal-Maine Farms, Inc., Cargill, Inc., Cargill Turkey Products, LLC, Georges, Inc., George's Farms, Inc., Peterson Farms, Inc., Simmons Foods, Inc., and Willowbrook Foods, Inc.
Defendants.

CASE NO. 05-CV-329-GFK-SAJ

in the United Station District Court
for the Northern District of Oklahoma

Expert Report

of

Roger L. Olsen, Ph.D.
CDM
555 17th Street, Suite 1100
Denver, CO 80202

Exhibit Q

Contents

Section 1 Introduction

Section 2 Sample Collection

Exhibit Q

Section 3 Laboratory Analyses

Appendices

Tables

Figures

Section 1
Introduction

This expert report provides my opinions and the supporting information for the following areas:

- Sample Collection (Section 2)
- Laboratory Analyses (Section 3)
- Database Compilation (Section 4)
- Laboratory Results (Section 5)
- Evaluation of Source of Contamination in the IRW (Section 6)

My opinions are stated below:

- The sampling approaches including selection of sampling locations and sampling methods were appropriate and resulted in collection of representative samples from the each major pathway environmental component.

- The sampling approaches and parameters selected for analyses are appropriate to identify all major sources and causes of contamination in the IRW environment including evaluations of impacts from cattle waste, poultry waste, and wastewater treatment plant (WWTP) effluent.

- The sampling approaches and parameters selected for analyses are appropriate to support the evaluation of injuries in the IRW.

- The analytical procedures selected for each parameter are appropriate for the intended data use (e.g., source identification and injury evaluation) and consistent with recommended methods by federal and state agencies.

- Except as qualified, the laboratory data are accurate, precise, representative, and comparable and can be used for all intended purposes and evaluations. The U.S. Environmental Protection Agency (EPA) recommended completeness goal of over 90 percent was achieved (98 percent complete) and this indicates that the data are of acceptable quality and can be used for its intended purposes.

- The stated objectives and hypotheses for the evaluation of sources of contamination in the IRW are appropriate to determine the effects and injuries resulting from poultry waste land application and other major sources of contamination in the IRW.

- The chemical and bacterial contaminants of poultry waste are found in all the environmental fate and transport components through out the IRW starting at the source of contamination (poultry waste disposal on fields) and including runoff water from the fields with poultry waste, surface waters, groundwater, springs, Lake Tenkiller water, river sediments and Lake Tenkiller sediments. Because the poultry related contaminants are pervasive through out the IRW, the overall water quality characteristics of the surface waters in the IRW have been substantially changed when compared to surface water quality in reference locations.

- The chemical and bacterial concentrations in each environmental component are consistent with known fate and transport processes and show a gradient in concentrations from high to low across the IRW depending upon closeness to poultry land application fields. These observations document a complete pathway of the poultry waste contamination from the land applied fields to streams, groundwater, springs, sediments, and Tenkiller water and sediments.

- The chemical compositions of the poultry waste and cattle manure are different from each other and individually unique. In addition, the chemical compositions of leachates of the poultry waste and cattle manure generated using synthetic precipitation are different from each other and individually unique. The chemical and bacterial compositions of poultry waste leachates are different and unique compared to WWTP discharges in the IRW. These differences allow identification of the important sources of contamination in the basin.

- Principal component analysis (PCA) identified two major sources of contamination in the IRW: poultry waste disposal and WWTP discharges. Poultry waste is by far the dominant contamination source in the IRW when compared to other sources. Cattle waste contamination was unique from both poultry waste and WWTP effluent and was identified in some samples with documented cattle manure contamination. However, chemical contamination from cattle waste is not dominant in the basin and only represents a minor source. In the PCA, the chemical and bacterial composition of poultry waste creates a distinct chemical signature that contains both phosphorus and bacteria.

- Mass balance calculations performed using the results of the synthetic precipitation leachates show that cattle manure is a relatively small source of the chemical contamination compared to poultry waste.

- Multiple lines of evidence by other experts (Drs. Engel, Fisher, Teaf and Harwood) support the conclusions that poultry waste is a major source of phosphorus and bacteria contamination in the IRW.

The information and evaluations supporting each of these opinions is provided in the following sections. Other opinions are also included in each section.

Section 2
Sample Collection

2.1 Poultry Waste

Application of poultry waste (litter) to fields has been identified as a probable source of environmental contaminants impacting surface water bodies, stream and lake sediments. The most notable environmental contaminant associated with this practice is phosphorous. Other contaminants include estrogens, bacteria, arsenic, nitrogen, copper, zinc, and other chemicals. The purpose of this task was to determine the constituents and concentrations in the poultry waste material. In order to determine if the constituents of poultry waste have been transported to the environmental components of the IRW and caused natural resources injuries.

2.1.1 Environmental Component

The environmental component is representative poultry waste from houses of the different poultry companies in the IRW.

2.1.2 Sampling Objectives and Intended Data Use

The purpose of this task was to characterize the bacterial and chemical constituents in poultry waste in order to evaluate and document links and relations between the poultry waste application and the environmental contaminants and conditions observed in the surface soils, surface water bodies, streams, groundwater and sediments of the IRW.

2.1.3 Type of Data to be Collected

Data collected during this field program consisted of bacterial and chemical composition samples of poultry waste.

2.1.4 Sampling Approach/Scheme

The sampling approach was to collect representative poultry waste samples from the major poultry producers in the watershed. In 2006 grower houses associated with the poultry producers Tyson, George's, Cargill, Petersons, Simmons, and Cobb were identified within the Oklahoma portion of the watershed and access obtained through a court ordered subpoena. Houses selected for sampling were proportionally selected from each poultry producer based on the approximate number of facilities each poultry producers was currently operating in the IRW. A majority of poultry waste samples were collected from houses containing broilers; however two houses were used to raise pullets, and 2 houses were used to raise turkeys. Additional houses were sampled in 2007 in order to provide a larger population for evaluation.

The sampling methodology is documented in the Standard Operating Procedures (SOPs) applicable to soil and litter sampling program. The SOPs are presented in Darren Brown's Expert Report (Brown 2008). The SOPs applicable to this sampling program are SOP 5-1 (Soil and Litter Sampling) and SOP 5-2 (Litter and Soil Sample

Compositing). In order to collect representative samples from the poultry house, CDM relied on the following documents in the development of SOP 5-1.

Zhang, H., Hamilton, D. W. and Britton, J. G. 2002. Sampling Animal Manure. Oklahoma State University Cooperative Extension Service Fact Sheet F-2248. Available at http://osuextra.okstate.edu/pdfs/F-2248web.pdf

Eucha/Spavinaw Watershed Management Team. Undated. Steps for Pulling Litter Samples.

Sampling staff utilized the zig-zag pattern similar to that identified in the above references in order to obtain a representative sample of the poultry waste. Zhang, et al. (2002) indicates that between 15 and 20 samples should be collected. The second reference (Eucha/Spavinaw Watershed Management Team) indicates that the poultry house should be divided into three longitudinal sections. Each section should have six samples collected, with a target of two from the feeding station areas, two from the water station areas, and two between the walls and the feeding/watering stations. The second reference (Eucha/Spavinaw Watershed Management Team) indicates that a total of 18 samples should be collected. Staff used the approach of Eucha/Spavinaw Watershed Management Team and collected approximately six samples around the water stations, six samples around the feeders and the remaining six samples were split between the walls and centerline of the house.

At each of the locations, a shovel was used to sample the entire depth of the waste and bedding material. Similar quantities from each sample location were placed in a five gallon bucket during the sampling.

The collected material was initially composited by mixing completely from top to bottom within a five gallon bucket to obtain a representative sample to be submitted for bacterial analyses. The remaining material was sent to the CDM Denver Laboratory for a more thorough compositing effort prior to submittal for the remaining analyses as described in CDM SOP 5-2.

2.1.5 Times to be Sampled

Poultry waste samples were collected in 2006 and 2007. The poultry house sample dates are provided in **Table 2.1-1**. With the exception of the Barnes' poultry house, sampling times of the poultry houses were dictated by the integrators and growers. Sampling was conducted between flocks, i.e., after one flock had been removed and before the next flock was placed in the house.

2.1.6 Field and Laboratory Analyses

No field parameters were collected during the sample collection. A composited portion of the samples were shipped directly to an offsite laboratory for the bacteria analysis with the remainder going to the CDM Denver laboratory for additional compositing prior to shipment to the remaining laboratories for various analyses. The analyses to be conducted on the poultry waste samples are documented in the SOPs applicable to soil and litter sampling program (SOP 5-1 and 5-2).

2.1.7 Implementation of Sampling Approach

The sampling methodologies are documented in SOPs 5-1 and 5-2 and are discussed in Darren Brown's Expert Report (Brown 2008) in relation to implementation. The field crews consisted of one staff member collecting the samples, one staff member providing videotape monitoring, and one or two staff members located off the property to provide decontamination support. As noted previously, timing of the sampling events was dictated by the integrators and growers and sampling was conducted while the flocks were not in the poultry houses with the exception of Barney Barnes' poultry house.

Sampling tools for this program consisted of a five gallon bucket, a short-handle spade, and a trowel. These sampling tools were purchased from a hardware store and used only once. The tools were either donated to the grower or thrown away after use. This was done to limit cross contamination issues as well as limit the possibility of disease transmission between poultry houses. Personnel protective gear was also disposed of between poultry houses and the footwear decontaminated with a soapy water rinse and scrub, followed by a ten percent bleach solution rinse, and then a tap water rinse. The decontamination effort was designed to not only limit the possibility of cross contamination between poultry waste samples, but to meet the requirements of the biosecurity protocols required by the integrators and Oklahoma Department of Agriculture, Food and Forestry (ODAFF). Additional information on the biosecurity protocols followed by CDM during poultry house sampling can be found in CDM SOP 5-2.

2.1.8 Alterations to the Sampling Program

In a small number of incidences, one less or a few more than 18 subsamples were collected and composited; however, in all cases the sample locations within each poultry house followed the general location guidelines as described in Section 2.1.4. The net result is that the number of collected samples provided a representative sample of the poultry waste; except for samples from FAC06. Because the inside of the poultry house was dark, some of the samples contained soil from below the bedding material.

2.1.9 Samples

A total of 20 poultry houses were sampled. **Table 2.1-1** provides a summary of the houses sampled by integrator, grower, and type of poultry in the house, sample ID, and sample date. The locations of the sampled houses are shown on **Figure 2.1-1**. In addition; some poultry waste samples (five) were collected as grab samples from the edge of the road where the poultry waste had fallen from trucks. These samples were collected before access was obtained to collect samples from poultry houses.

2.2 Soil

2.2.1 Environmental Component

The environmental component is soils from fields where poultry waste has been applied. In addition, soils were also collected from fields with no reported poultry waste application (see Section 2.13 for comparison). Soils from fields with poultry waste application represent the first environment component.

2.2.2 Sampling Objectives

The purpose of this task was to characterize bacterial and chemical constituents in soils in fields where poultry waste has been applied in order to evaluate and document links and relations between the poultry waste land application and the environmental contaminants and conditions observed in the surface water bodies, streams, groundwater and sediments of the IRW.

2.2.3 Type of Data to be Collected and Intended Use

Data collected during this field program consisted of the bacterial and chemical composition samples of surface soils from the zero to six-inch depth. The soil samples were collected from fields with known poultry waste application as well as fields without known poultry waste application (see Section 2.13). In addition to the field data, land application records were obtained from the Oklahoma Department of Agriculture, Food, and Forestry (ODAFF) to determine which field has received poultry waste application.

Soil samples were collected from the zero to 2-inch, 2- to 4-inch, and 4- to 6-inch depths from sampled fields. The collection of soil from three distinct depth intervals facilitates the understanding of how surface application impacts the soils within an applied field. It provides a more detailed understanding of the fate and transport processes associated with poultry waste application. The zero to 2-inch sample represented more closely soil that may runoff the fields during precipitation events. The zero to 2-inch sample was analyzed for a larger list of parameters than the lower two depths.

The analytical data from the zero to 2-inch sample were used to determine a complete bacterial and chemical composition of the soil. The same parameters were analyzed in poultry waste and soils. All samples were analyzed for Mehlich Phosphorus (P) in order to provide a direct comparison of the data with the historical ODAFF data for the grower's properties, which provided Mehlich P in a zero to 6-inch soil sample.

2.2.4 Sampling Approach/ Scheme

The goal of this sampling program was to collect representative soil samples from both poultry waste applied fields and non-applied fields (see Section 2.13).

Prior to the 2006 soil sampling program, properties were identified as potential candidates for soil sampling based initially on available ODAFF Animal Waste Management Plan records which showed that poultry waste had been applied to the fields in at least 3 of the previous 5 years. Sites were selected so that properties upon which poultry waste had been applied from each of the major poultry producers within the watershed were sampled. All properties selected for soil sampling required a subpoena from the Court to allow CDM access to sample the property, with the exception of Barney Barnes' property, who voluntarily allowed CDM to sample. In some cases, the originally selected property was replaced by an alternate property due to changes in land ownership prior to sampling. The 2007 soil sampling locations were again selected based on review of available ODAFF Waste Management Plan reports. In addition, several fields were sampled in 2006 and 2007 which had not previously been applied with poultry waste. These control fields are described in greater detail in Section 2.13 of this report.

In general, four sampling grids were established for each grower's property. Each sampling grid was typically between one and ten acres, depending upon the field size. The grid was typically established for an area that was generally uniform in nature with respect to topography and soil type and was treated as one waste application field with respect to the ODAFF Animal Waste Management Plan records or Nutrient Management Plan records. Each sampling grid typically consisted of twenty sampling stations. SOP 5-1 established the number of sampling stations as twenty per sample grid based upon the information provided in Appendix A-3 of SOP 5-1. Appendix A-3 of SOP 5-1 provides reference to work conducted by Oklahoma State University and other universities which describe how to collect a representative composite sample from soil cores within a field. The graph provided shows the variability is significantly reduced after twelve composited samples, but twenty is considered optimal. The grid spacing developed by CDM was a four node point by five node point grid. A regular grid (versus random locations) eliminates any potential bias by staff conducting the soil sampling and maximizes the amount of information provided by a given number of samples. The sampling plan did allow for node points to be adjusted where certain surface features were present that could potentially bias the sample results. Per the instructions of Appendices A-3 and A-4 of SOP 5-1, the node points could be adjusted to avoid sample collection in unusual spots that are not typical of the field as a whole. Examples of unusual spots include wet spots, low spots, livestock feeding and loafing areas (unless a significant part of the field). CDM SOP 5-1 also included the provision to avoid sampling under the drip line of trees if possible although this advice was not set forth in Appendices A-3 or A-4 of SOP 5-1.

Each sampling station was typically subject to between one and three sample cores advanced to a depth of six or more inches. CDM identified the surface to two-inch soil sample as the interval likely to be most impacted by poultry waste application due to regional soil characteristics. Additionally, the surface to two-inch interval is expected to be the most likely soil zone to contribute nutrients and sediment to the watershed during significant runoff/erosion events. Therefore, most of the zero to two-inch soil samples were submitted for full suite (list of parameters) analysis (nutrients, metals, bacteria, and estrogens) as identified in the analysis text below. A partial suite of analyses consisting of nutrients and metals analyses was conducted on most two to four-inch soil samples.

For the purposes of this investigation, complete decontamination of personnel and sampling materials only needed to occur when personnel and equipment were leaving a subject property onto a public right-of-way. Complete decontamination as detailed in SOP 5-1 would address the biosecurity concerns of the growers, integrators, and ODAFF and significantly minimize the potential of cross contamination between different grower's properties. Decontamination between individual sample depths, cores at a grid node point, or between grid node points within a sample grid was not specified in SOP 5-1. Decontamination between sample grid node points is not discussed in Appendices A-3 or A-4 of SOP 5-1 because the material is composited into one sample. Decontamination between sample depths is not necessary because the volume of potential cross contamination is minimal relative to the overall sample volume non-uniform nature of soil and the analytical tests being performed (see further discussion in Section 3). For the same reason, decontamination between sample cores and sample node points is not likely to provide a measurable or significant impact. Impact in these cases is defined as data that would change enough to effect the intended data use.

SOP 5-1 does indicate that decontamination of the sampling materials (corers, knife, and ruler) should take place when moving from one sample grid to another within a grower's property and not crossing public right-of-ways. The decontamination process was removing any excess dirt from the sampling equipment prior to continuing the sample program. This was accomplished by scraping the soil off and then driving and removing a pilot core (at the new grid location) between sampling of each sample grid within a property. All sampling equipment was fully decontaminated between properties by removing any visible soil and rinsing the equipment with phosphate-free detergent, distilled water, and a 10% bleach solution. In addition, soil corers were power-washed with clean tap water between each property.

2.2.5 Times to be Sampled

Soil samples were collected in 2006, 2007, and 2008. The soil sample dates are provided in **Table 2.2-1**. Soil samples were not collected during or immediately following precipitation events of a magnitude capable of fully saturating the top 2 inches of soil at a specific sampling location.

2.2.6 Field and Laboratory Analyses

No field parameters were collected during the sample collection. The soil samples were shipped to the CDM Denver laboratory for additional compositing prior to shipment to the analytical laboratories for various analyses. The analyses to be conducted on the soil samples are documented in Standard Operating Procedures (SOPs) 5-1 (Soil and Litter Sampling) and SOP 5-2 (Litter and Soil Sample Compositing). The SOPs are presented in Darren Brown's Expert Report (Brown 2008).

2.2.7 Implementation of Sampling Approach

The sampling methodology is documented in the SOPs applicable to soil and poultry waste sampling program. As discussed above, the SOPs applicable to this sampling program are SOP 5-1 and SOP 5-2. All samples were labeled and bagged individually prior to shipment to the CDM Denver laboratory. Decontamination procedures were consistent with SOP 5-1. All procedures were conducted according to the applicable SOP except as discussed in the following section.

2.2.8 Alterations to the Sampling Program

In some cases, property sizes were insufficiently large to establish four separate sample grids so only 2-3 sample grids were established on that property. In other instances, rocky soils or shallow bedrock interfered with the ability to collect soil material from select target depths. Sample core recoveries were noted in the field books used to document sampling efforts. Recoveries were rated as "good", "moderate", "poor", and "no recovery". "Good" recovery meant a full sample recovery for the interval in question. "Moderate" recovery indicated a recovery of between fifty percent and full recovery. "Poor" recovery indicated a recovery of some sample volume between zero and fifty percent. In most cases, "moderate" and "poor" recovery efforts were due to the presence of pebble-sized material which was physically removed prior to placement of the soil sample in the sample bag.

In addition to the Litter Applied Locations (LALs) and Control Locations (CLs) previously discussed, another soil sampling program was implemented in 2008. In this instance two fields used to pasture beef cattle but not previously applied with poultry waste were sampled using three grids. The pastures were designated as Cattle Pastures with a CP designation. Sampling was conducted according to the SOPs.

In general, sample grids were collected from individual waste application fields. In some cases where the fields were significant in size and/or the number of individual application fields associated a grower was limited, more than one sample grid might be established within a waste application field. **Figure 2.2-1** shows the general location of each grower property sampled during this program with LAL, CL, and CP IDs included. The positions of each sample grid relative to the grower's property are shown on **Figures 2.2-2** through **2.2-28**.

2.2.9 Samples

Including the non-litter applied fields discussed in Section 2.13, a total of 86 grids were established and sampled during this field program. Sample grids were established on the properties of sixteen separate growers and six non-growers where poultry waste had been applied. **Table 2.2-1** provides a summary of the sample grids by integrator, grower, sample ID, and sample date. As noted above, sample locations are provided on **Figure 2.2-1**, and **Figures 2.2-2** through **2.2-22**.

2.3 Edge of Field (Surface Runoff Water)

2.3.1 Environmental Component

Edge of field sampling following precipitation events was established to collect representative samples of surface runoff water from fields of which poultry waste was recently applied.

2.3.2 Sampling Objectives

The primary sampling objective for edge of field sampling was to obtain samples of surface runoff water from fields to which poultry waste had been recently disposed in order to evaluate and document links and relations, if any, between poultry wastes land application and environmental contamination within the IRW.

2.3.3 Type of Data Collected and Intended Use

Edge of Field runoff water was collected and sent to a variety of laboratories for analyses of a full suite of chemical parameters, including nutrients and metals, as well as a number of biological parameters such as bacteria. At the time of sampling, additional field data were collected pertaining to the exact location of the sampling point and relative direction of runoff flow. The data obtained from these surface runoff samples provided information concerning the concentrations of nutrients, metals and bacteria in runoff water that had interacted with poultry waste that had recently been disposed by land application. These data were intended to provide information on the transport of contaminants in poultry waste from applied fields to surface water bodies and to groundwater.

2.3.4 Sampling Approach

Fields to which poultry waste had recently been applied were identified and sampled following one of two methods. Initially, potential sampling locations where identified either by direct observation of poultry waste disposal or through the observation of indications of recent poultry waste disposal (tire track pattern, damaged vegetation, patterns of vegetative growth, poultry feathers and waste on the surface). Once a potential location was identified, field crews located the apparent lowest point of the applied field where runoff would be expected to flow. In cases where the field appeared to drain to an accessible right-of-way, field crews would install a small, approximately one gallon, PVC collection tube in a location likely to collect the runoff. The tube was installed into the ground so the top was at ground surface. During or immediately following a significant rain event in the area, CDM or Lithochimeia personnel would return to the site to sample any water which had accumulated in the PVC collection tube.

In 2006 and 2007, direct observations of poultry waste application or indications thereof, were documented by private investigators contracted to canvass the watershed to document the commercial application of poultry waste to fields. Direct observation of poultry waste applications by the private investigators began in March of 2005 and continued through the spring of 2007. Typically, 2 teams of investigators were in the basin for several days each week and during peak application periods, as many as 4 separate teams of investigators were employed simultaneously. Often, the investigators would observe a fully loaded application vehicle (spreading truck) and follow it to the intended application field. The investigators would then document the application by filling out a field notes providing field description, time and date of application, and by photographing or videotaping the application. A GPS coordinate was recorded and typically a general sketch of the location was created noting the location of and potential access to the apparent lowest point where runoff would be expected to flow.

Field observation notes from the investigators were supplied to CDM or Lithochimeia employees via email or through verbal communication on a daily basis. Because a significant rain event was required to collect runoff from the recently applied locations, CDM and Lithochimeia field crews monitored the local weather reports in order to anticipate the timing of potential sample collection events. The intention was to be onsite at a recently applied location during or immediately after a precipitation event large enough to create runoff for sample collection. Samples were collected next to fields, ditches, culverts, and drainage basins coming directly off of applied fields.

2.3.5 Times to be Sampled

Samples were collected during or as soon following a runoff producing rainfall event as possible. Runoff samples were typically collected between 1-12 hours of when precipitation began.

2.3.6 Field and Lab Analyses

Edge of field runoff samples were analyzed for nutrients, certain organic compounds, metals, and bacteria. In 2006 and 2007, bacteria and phosphorus samples were sent directly from field to EML and Aquatic Research laboratories for analysis.

2.3.7 Changes to Sampling Scheme

Edge of field samples between 5-14-2005 and 6-05-2005 were taken either using PVC sample capture tubes buried at locations where runoff was predicted to exit fields or on a several occasions, collected as direct grab samples when run-off from land applied fields was observed. Because it proved difficult to reliably identify locations where sufficient runoff volume could be collected, all samples collected in 2006 and 2007 were collected by directly placing runoff water into sample containers. A small sterilized bucket and/or funnel were used to collect samples into (1-2) 1 gallon sterile jugs and bacteria samples were collected in (2) 250 mL or (1) 500 mL sterile bottle and sent directly to the lab oratory. All equipment that was reused was scrubbed with phosphate free detergent and rinsed 3 times with deionized water.

Originally samples collected were field filtered and otherwise prepared and shipped directly to appropriate laboratories. To facilitate processing and filtering, a decision was made to send bacteria and phosphorus samples directly to appropriate laboratories and the remaining volume to the CDM Laboratory in Denver for sample preparation and shipment to other laboratories. Bacteria and phosphorus samples were shipped via overnight service directly to the laboratories on the day of collection.

2.3.8 Sampling Summary

There were a total of 89 Edge of Field samples collected. The locations of all edge of field samples collected during the course of this program are shown in **Figure 2.3-1**. A summary of the total number of edge of field samples, sample dates, and analyses is provided in **Table 2.3-1**.

2.4 Small Tributary Sampling

2.4.1 Environmental Component

The environmental component targeted for the Small Tributary Sampling in 2005 and 2006 was the surface water in small sub-basins and associated tributaries of the Illinois River.

2.4.2 Sampling Objectives

The overall purposes of the water quality investigations in the IRW were to evaluate and document the links and relations, (if any) between the poultry waste land application and environmental contamination within the IRW. Another objective of this program was to directly quantify contaminant loads and concentrations in upstream (smaller watershed) tributaries, during both elevated and base flow conditions, for a range of sub-basin poultry activity levels.

2.4.3 Type of Data Collected and Intended Use

Small upstream tributaries may represent a direct link between watershed characteristics and activities and downstream receiving water quality. These tributaries are highly sensitive to both the hydrology and contaminant loadings associated with the contributing drainage area. Local groundwater inflows and loadings impact tributary baseflow quality and quantity. If large and intense enough, precipitation events will result in water runoff and contaminant loadings from the drainage areas to the tributaries. Most of the transport of sediment and suspended matter in the streams and from the landscape occurs during runoff events associated with precipitation or storms. Therefore, sampling of upstream tributaries in the watershed during elevated flow is important for quantifying total loads to downstream receiving waters. Additionally, by focusing on upstream tributaries, many confounding factors can be removed from the analysis, such as instream processing and point source loadings, and the relationship between poultry presence and stream water quality can be isolated.

Data collected during this sampling program included a full suite of chemical parameters, including nutrients and metals, as well as a number of biological parameters such as bacteria. Hydrologic conditions at the specific time and location of each sample collection event was also documented. Additional land-use data for each sub-watershed was also collected and analyzed during the course of this program.

The data generated from this study were used to establish empirical links between stream water quality and a variety of land-use characteristics, including poultry operators presence. These empirical links are used in a predictive model to calculate "event mean concentrations" and baseflow concentrations as a function of poultry house densities. With these concentrations, simple hydrologic calculations, and known point source loadings, the model is able to predict sub-basin phosphorus loads across the watershed for a range of long-term flow scenarios. Finally, the data were used to calibrate a mechanistic rainfall-runoff model of the entire watershed that makes similar predictions using a series of process-based equations. These analyses are all described in Section 6.9 and in Engel's Expert Report (Engel 2008).

2.4.4 Sampling Approach/ Scheme

2.4.4.1 Methods: Stratified Design

This sampling program measured concentrations and flow characteristics during high flow events and baseflow periods, using automated samplers, at twelve locations within the IRW. The sampling was performed during spring and summer months of 2005 and 2006. Two of the locations were changed from 2005 to 2006, as described below. The sampling locations were all in lower order streams (second and third order) draining sub-basin areas ranging from 1 to 41 square miles. The program was coordinated with the United States Geological Survey (USGS) sampling of the higher order streams (third, fourth, fifth and sixth order streams) at established sampling locations that represent much larger watersheds. The number of sampling locations utilized in this study (twelve per year) was set to be sufficient to a) achieve significance in subsequent statistical analyses and b) cover a range of basin characteristics, with respect to the criteria defined below; that is, representative of the range seen throughout the entire watershed.

Sampling locations were selected based on the following characteristics:

- Poultry house density (number of houses in the sub-basin being sampled, a surrogate for potential contamination)
- Stream order and flow
- Proximity to USGS flow gages (available data)
- Land use (poultry waste application fields, pasture, and forest)
- Geographic distribution (adequate spatial distribution across the watershed).

Poultry house densities were determined from a combination of aerial photographs and field reconnaissance (see Section 2.15). Initially, 27 potential sampling sub-basins across the watershed were chosen that met the size criteria (approximately 1 to 41 square miles) and the sampling locations appeared to be accessible based on map inspection. Poultry house densities were calculated for each, and the range of these densities was divided into five equal sub-ranges (quintiles). At the next screening level, three to four sites within each quintile were selected as potential sampling sites. Finally, field inspection narrowed these 27 locations down to the final twelve sites for each year based on accessibility and the ability to secure the automated samplers (described below), while maintaining two to three sites within each density quintile.

Sampling stations were located at the downgradient end of sub-basins to collect runoff and mass loadings associated with upgradient drainage areas. As described above, the poultry house densities in each of the selected sub-basins are representative of the range of house densities found in the IRW, including at least two "reference" sub-basins with minimal or no poultry presence. The densities of active chicken houses in the sub-basins sampled ranged from zero to approximately 15 houses per square mile. Sample site locations are shown in **Figures 2.4-1a** and **2.4-1b** and summarized in **Table 2.4-1**. Note: In table 2.4.1, site 10 14 (HFS14) shows zero poultry house density. After the program started, poultry waste disposal was observed near the river where the sampling station was located.

2.4.4.2 Methods: Sample Collection

ISCO Automated Samplers (Model 6712) were used for all sampling events. These samplers draw samples from a given stream according to pre-programmed specifications and can be triggered by increases in stream stage height or flow rate. They are widely used and accepted by the scientific community for this type of application.

The automated samplers were each housed in drum-like containers approximately 20 inches in diameter by 30 inches high. **Figure 2.4-2** shows a photograph of a sampler deployment and a schematic of a typical deployment configuration. Each sampler collected 24 discrete 1-liter samples of stream water in polyethylene bottles for each highflow event. Compositing of these samples is described in Section 3.5. For the baseflow events, the required sample volumes were collected by the samplers and no compositing was performed. During sampling, streamwater is pumped through a suction line with a peristaltic pump housed in the top of the drum, and is distributed to the bottles via a rotating arm. Each sampler was equipped with an ISCO 750 Flow Meter that monitored stream stage height and velocity. Each sampler was programmed to initiate highflow sampling when a threshold stream stage was reached. The threshold stage was set specific to each stream based on known storm hydrograph characteristics. The value aimed to be low enough to capture, to the extent possible, the full hydrograph of a given storm event, but also high enough to only sample significant runoff events. In some cases this value was determined by trial and error at the beginning of the sampling season

Sampling intervals for a given storm event, for all sites in 2005 and most sites in 2006, were every one hour for the first ten hours and every three hours for the subsequent 42 hours (for a total of 24 samples over a 52 hour period). For the 2006 field season, the sampling intervals at four of the smaller, high gradient tributaries (HFS05, HFS21, HFS28A, and HFS30) was altered due to the shorter duration of precipitation impacts on these streams. The sampling intervals for these streams were reduced to every 0.5 hours for first 2 hours (five samples), and every 2 hours for remaining samples (total of 40 hrs for full hydrograph). The goal of this sampling regime was to collect a representative distribution of samples during both the rising limb (shorter) and the falling limb (longer) of the stream hydrograph. The design of this regime was guided by gaged flow data for small tributaries in the watershed in response to storm events. An example hydrograph with sample collection points is shown in **Figure 2.4-3**.

2.4.4.3 Methods: Laboratory Analysis

Immediately following each sampling event, discrete bottle samples were collected by a field team, placed in a cooler on ice, and shipped to CDM Denver Laboratory for sample preparation.

For high flow events, collected samples from a given site were composited, using flow weighting techniques, into a single sample representative of the entire event hydrograph. This is standard procedure for measuring so-called "event mean concentrations" of runoff events. Flow weightings were based on either measured stream depths or, when adequate velocity data were available, flow estimates from measured depths and velocities. Relative weightings were assigned to each discrete sample based on the area under the event depth or flow curve for the discrete sampling interval as compared to the total area under the curve. In other words, samples collected during the peak of the hydrograph carried more weight than those collected near the bottom of the hydrograph, given the same sampling interval. Likewise, samples collected for a 3 hour interval carried more weight than those collected for a 1 hour interval, given similar flow rates. In addition to the composited samples, portions of a number of peak flow discrete samples were retained for laboratory analysis to compare discrete peak flow concentrations to event mean concentrations and to gain information on the variability in concentrations during the event. Baseflow samples were not composited.

In 2005, composite samples were sent to EML for analyses of bacteria. In 2006, grab samples collected near peak flow were sent directly to EML from the field for bacteria analysis.

2.4.5 Sample Times

The ISCO Automated Samplers were initially installed the week of 4-13-2005 and all 2005 samples were collected between 5-25-3005 and 10-12-2005. All samples during 2006 were collected between 3-9-2006 and 8-1-2006.

2.4.6 Field and Laboratory Analyses

Following compositing at the CDM Laboratory, samples were submitted to analytical laboratories for various analyses including nutrients, metals, bacteria, and estrogens. Details of these analyses are provided in **Table 2.4-2**. As shown, both total (non-filtered) and dissolved (filtered) metals and phosphorous were analyzed. For dissolved metals and phosphorous, the samples were filtered using a 0.45 micron filter. Stream stage height and velocity field data were collected by each sampler equipped with an ISCO 750 Flow Meter. Field crews typically collected water temperature, pH, and conductivity data on site during the time of sample retrieval. In addition, field measurements of stream turbidity and dissolved oxygen were typically collected at each site during baseflow sampling.